

Influenza Virus Testing at the Washington State Public Health Laboratories (WAPHL) August 23, 2010

The Washington State Public Health Laboratories (WAPHL) primarily perform influenza virus subtyping and oseltamivir resistance testing for surveillance purposes. Results are used to monitor influenza activity in the state and guide influenza treatment recommendations. Subtyping is performed using a real-time reverse transcriptase polymerase chain reaction (RT-PCR) test developed by the Centers for Disease Control and Prevention. Oseltamivir resistance testing is performed by screening for the H275Y mutation in the neuraminidase gene using pyrosequencing.

After approval from the local health jurisdiction, WAPHL will perform influenza testing and subtyping on specimens from:

1. Deceased patients suspected to have influenza (**NOTE:** Autopsy specimens are not an approved specimen type and thus viral isolation must be performed prior to performing RT-PCR testing for characterization).
2. Patients suspected to be infected with a novel strain, including H5N1 influenza.
3. Patients associated with outbreaks.

After approval from the local health jurisdiction, WAPHL will perform oseltamivir resistance testing for clinical care purposes on specimens from:

1. Patients who develop laboratory-confirmed influenza while taking antiviral prophylaxis.
2. Severely immunocompromised patients with prolonged excretion of influenza virus despite antiviral treatment.
3. Patients in intensive care units with prolonged excretion of influenza virus despite antiviral treatment.

In addition, WAPHL will perform subtyping and oseltamivir resistance testing on specimens from ILI Net sentinel providers and sentinel laboratories around the state. These results will be published weekly in the DOH Influenza Update (<http://www.doh.wa.gov/ehsphi/Epidemiology/CD/fluupdate.pdf>).

Specimen collection and storage

The following specimen types are acceptable for influenza testing at WAPHL:

- Nasopharyngeal swab
- Nasal aspirate or wash
- Throat swab
- Nasopharyngeal / Throat swab
- Tracheal aspirate
- Nasal swab
- Viral culture
- Bronchoalveolar lavage (BAL)
- Bronchial aspirate or wash

Healthcare providers should:

- Collect specimens using appropriate infection control procedures.
- Collect nasopharyngeal and throat swabs using swabs with a synthetic tip, such as Dacron or nylon, and a plastic or wire shaft. Specimens collected with cotton or calcium alginate swabs with wooden shafts will not be tested.
- Immediately after collection, place the swab or aspirate material into a sterile vial with 2 ml of viral transport media; for swab specimens, break or cut off the end of the swab shaft.

QUESTIONS? Most questions should be directed to your local health jurisdiction.
Communicable Diseases Epidemiology may be reached at (206)418-5500
WAPHL, Virology Laboratory may be reached at (206)418-5458

- **Close the vial tightly** to avoid leakage during transport. Specimens that leak in transit will not be tested at WAPHL.
- Do not let a swab come into contact with reagents used for other tests. If a swab contacts reagents for other tests, a new swab must be submitted.
- Label the vial with the patient's name and a second identifier, specimen source, and date obtained; place immediately in a refrigerator (2 - 8°C) until ready for transport. **DO NOT FREEZE.**

Specimen shipping

Please Note: Specimens must be received at the Public Health Laboratories (WAPHL) within 72 hours of collection. Specimens which arrive more than 72 hours after collection cannot be tested using RT-PCR. WAPHL is open to receive influenza specimens Monday through Friday 8 am–5 pm.

- Complete an Influenza Testing Specimen Submission Form, available at: <http://www.doh.wa.gov/EHSPHL/PHL/Forms/SerVirHIV.pdf>. **Specimens will not be processed until ALL of the following information is known:**
 - Patient name and county of residence
 - Specimen type and date of collection
 - Submitter contact information
 - Reason for testing at WAPHL (i.e., which criteria are met by the patient)
- Make sure the name and date on the vial match the name and date on the specimen submission form.
- Place the completed Influenza Testing Specimen Submission Form into the OUTER pouch of the plastic biohazard envelope. Do not place any paperwork in the inner pouch along with the vial.
- Pack and label clinical specimens as Biological Substances, UN 3373. Pack and ship according to United States Department of Transportation and United States Postal Service regulations.
- Transport clinical specimens cold (NOT frozen) on ice packs to:

Washington State Public Health Laboratories
Attn: **WAPHL** Virology Laboratory
1610 NE 150th Street
Shoreline, WA 98155
- Freeze viral isolates to -70 °C and ship on dry ice. Please contact the WAPHL Virology Laboratory for further instructions on shipping viral isolates: (206) 418-5447

WAPHL testing procedures

Test results turnaround time: Projected turnaround time for influenza testing and subtyping using RT-PCR or oseltamivir resistance testing will be 5 to 7 business days from receipt of the specimen at the WAPHL. If a specimen requires both influenza testing and oseltamivir resistance testing or additional work, such as viral isolation at the WAPHL or submission to the CDC for additional testing, then final results may be delayed.

Reporting of test results: Test results are sent by postal mail to the submitting facility. Test results are also sent by fax and/or electronic reporting system to the local health jurisdiction in which the patient resides.

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